ANTIBODIES IN THE CHICK

HAL W. SHERMAN

From the John McCormick Institute for Infectious Diseases, Chicago

Klemperer[1] found that diphtheria antitoxin was transmitted from immunized fowls to the vitellus of the egg. Dzierzgowski[2] obtained the same results, and in addition reported that the blood of chicks hatched from such eggs contained antitoxin. Figari[3] fed tubercle bacilli to hens and demonstrated specific agglutinins in the eggs; he found no agglutinins in control eggs.

The scope of my work was to study natural antibodies of hens, chicks, and chick embryos. Lysins for sheep, goat, rabbit and human corpuscles have been estimated; also complement. Blood was obtained from the wing vein of the hen, and the jugular veins of chicks. To get blood from the embryo without contamination by other fluids is more difficult. The incubated eggs were placed on a candling device and the shell and outer membrane over the air chamber removed. As the inner membrane came into view xylol was brushed carefully over it; the membrane then became transparent, the blood vessels stood out prominently, and one of the largest vessels could now be punctured with a needle and the embryo bled to death into the air chamber, from which the blood was removed with a pipet before clotting occurs. Great care was exercised not to go through the membrane with the needle, as then other fluids came out with the blood and air gained entrance into the egg substance with consequent bulging of the inner membrane and shell. Anywhere from ½-1 c c of blood could be obtained from an embryo in this way.

Several different batches of chick serums were tested at different times and for convenience the results are given in table 1. In no case was lysin found in any embryo except those of 21 days' incubation, and in this case the chicks were pecking their way out of the shell. Their serum contained lysin for rabbit erythrocytes only, 0.1 c c of serum being required to lake completely 0.1 c c of a 1% erythrocyte suspension. Complement was found in the embryo serums of 17 and

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[1] Arch. f. exper. Path. u. Pharmacol., 1893, 31, p. 356.
[2] Centralbl. f. allg. Path. u. path. Anat., 1901, 12, p. 715.
[3] Centralbl. f. Bakt., I, O., 1907, 39, p. 75.

21 days' incubation; it required 0.05 c c of the serum of the younger embryo and 0.1 c c of that of the older to lake completely 0.1 c c of a 1% suspension of rabbit erythrocytes when 1 unit of amboceptor (dog) was added. No embryo serum examined contained complement for sheep, goat or human erythrocytes. The embryo serums were not anticomplementary. Lysins and complement were not present in the amniotic or allantoic fluids or in fresh nonincubated eggs.

TABLE 1

RESULTS OF DIFFERENT TESTS

Age of Chick	Complement Titers Action of Serum + 1 Unit of Amboceptor on Erythrocytes of				Lysin Titers Action of Serum + 1 Unit of Complement (Guinea-pig Serum) on Erythrocytes of			
	Sheep (Rabbit Ambo-ceptor)	Goat (Rabbit Ambo-ceptor)	Human (Rabbit Ambo-ceptor)	Rabbit (Dog Ambo-ceptor)	Sheep	Goat	Human	Rabbit
Pecking at shell	no lysis	no lysis	no lysis	+++ (0.1)	no lysin	no lysin	no lysin	+++ (0.1)
1 hour	no lysis	no lysis		+++ (0.01)	no lysin	no lysin		+++
10 hours	no lysis	no lysis		+++	no lysin	no lysin		+++
24 hours	no lysis	no lysis		+++	no lysin	no lysin		+++
24 hours	no lysis	no lysis	no lysis	+++ (0.1)	+++ (0.2)	no lysin (0.3)	no lysin (0.3)	+++ (0.1)
24 hours	no lysis	no lysis		+++ (0.01)	+++ (0.05)	no lysin		+++
48 hours	+++	+++	+++	+++	+ (0.1)	+++ (0.1)	+++ (0.1)	+++
72 hours	+++	+++		+++ (0.01)	+++	no lysin		+++ (0.01)
21 days	+++ (0.1)	+++ (0.1)	+++ (0.1)	+++ (0.1)	+++ (0.1)	no lysin	+++ (0.1)	+++
Full grown	+++ (0.1)	+++	+++ (0.1)	+++	+++	+++	+++	+++

0.1 c c of a % erythrocyte suspension was used for the older chicken while in the newly hatched 0.1 c c of a 1% suspension was taken.

Unless otherwise stated by figures in parentheses, the amount of chick serum used was either 0.05 or 0.03 c c.

+++ means complete laking; ++ partial laking; + slight laking.

In a previous paper[4] I reported that lysins and complement are inappreciable in the youngest swine embryos; they were found in varying amounts, however, after the 9th week of gestation. Whether these antibodies were autochthonous or transmitted through the placenta from the maternal blood seems impossible to determine, although their detection only in the 9th week would seem to make more probable the view that they came through the placenta. Histologists have shown a less intimate relationship between the fetal and maternal blood vessels in early embryonic life than later. If this anatomic

[4] Jour. Infect. Dis., 1919, 24, p. 1.

difference permits of easier transmission of nutrition to the fetus, then why not also of antibodies? The study of the chick embryo was undertaken to avoid the complication of the transmission of antibodies from the maternal blood through the placenta. No antibodies were found in the chick embryos before the 21st day of incubation when all the embryos examined were pecking at the shell. This is an entirely different condition than that presented by the swine fetus. How are we to account for the lysins of the 21-day embryo and the lack of them in the 19-day embryo? There are two physiologic differences in the embryos of these ages. In the 19-day embryo respiration is maintained by the capillaries of the internal membrane exclusively, and in the 21-day embryo this capillary network is aided by occasional inspiration by the lungs; this is of doubtful significance. It is necessary to consider another factor. With the chick pecking on the shell, it seems possible that there may be a stimulation of secretion from gastro-intestinal glands with subsequent absorption of antibodies into the blood. Whether antibodies and complement occur in the gastro-intestinal secretions is a moot subject. R. Neumann[5] has described a dog in which the large intestine was cut off from the small intestine and the ends tied; the free end of the small intestine was sutured to an artificial anus; after several months the secretion from the large intestine was collected and found to contain hemolysins qualitatively the same as those of the blood. Whether lysins in the intestines were derived from the blood is a question; there may be no relationship, but this seems unlikely. Metchnikoff[6] believed that the fixatives of the blood serum were set free as leukocytes disintegrate. Is there an increased leukocytolysis during the 20th or 21st day of incubation that can account for the presence of lysins in the 21-day and not in the 19-day embryos? There was no increase in the lysin titers of the swine embryos[4] in the last days of intra-uterine development. The source of the lysin found in the hatching and newly hatched chick remains obscure.

[5] Arb. a. d. path. anat. Inst. zu Tübingen, 1911, 7, p. 546.
[6] Immunity in Infective Diseases, 1905, p. 98.